

CLAIMS

1. An *in vitro* method of screening human subjects for the presence of human papillomavirus in at least one cell or tissue, wherein the human papillomavirus exhibits loss of regulation of E6/E7 mRNA expression and loss of replication and/or expresses a stabilized pre-mRNA encoding full length E6 protein, the method comprising detecting the presence of mRNA transcripts of the E6/E7 gene of a human papillomavirus which encode full length E6 protein in a test sample comprising mRNA derived from the cell or tissue, wherein the presence of such E6/E7 mRNA transcripts in the sample is taken as an indication of the presence of human papilloma virus exhibiting loss of regulation of E6/E7 mRNA expression and loss of replication and/or expression of a stabilized pre-mRNA encoding full length E6 protein in the cell or tissue.

20

2. An *in vitro* method of screening human subjects for the presence of cellular changes characterized by enlarged cell nuclei and cellular aneuploidy in at least one cell or tissue, which method comprises detecting the presence of mRNA transcripts of the E6/E7 gene of human papillomavirus which encode full length E6 protein in a test sample comprising mRNA derived from the cell or tissue, wherein the presence of such E6/E7 mRNA transcripts in the sample is taken as an indication that the cell or tissue under test exhibits the cellular changes.

25

30

3. An *in vitro* method of screening human subjects for the presence of persistent transforming infection with human papillomavirus in at least one cell or tissue, which method comprises screening the subject for expression of mRNA transcripts of the E6/E7 gene of human papillomavirus which encode a full length E6 protein in a test sample comprising mRNA derived from the cell or tissue, wherein subjects

35

40

- 42 -

positive for expression of such mRNA transcripts of the E6/E7 gene of human papillomavirus are scored as having a persistent transforming infection with human papillomavirus in the cell or tissue.

5

4. A method according to any of claims 1 to 3 which comprises detecting the presence of mRNA transcripts of the E6/E7 gene of human papillomavirus using a technique which is able to detect E6/E7 mRNA from at least one cancer-associated HPV type.

10

5. A method according to claim 4 which comprises detecting the presence of mRNA transcripts of the E6/E7 gene of human papillomavirus using a technique which is able to detect E6/E7 mRNA from HPV types 16, 18, 31, 33, and preferably 45.

15

6. A method according to claim 4 which comprises detecting expression of mRNA transcripts of the E6/E7 gene from any one or any combination of two or more of HPV types 16, 18, 31, 33 or 45, wherein the presence of mRNA transcripts of the E6/E7 gene of human papillomavirus from any one of the tested HPV types in the sample is taken as a positive result.

25

7. A method according to any one of claims 1 to 6 wherein detection of expression of mRNA transcripts of the E6/E7 gene is carried out using an amplification reaction to amplify of a region of the mRNA, together with real-time detection of the products of the amplification reaction.

30

8. A method according to claim 7 wherein detection of expression of mRNA transcripts of the E6/E7 gene is carried out using real-time NASBA.

35

9. A method according to claim 8 wherein detection of expression of mRNA transcripts of the E6/E7 gene is carried out using the Pre-Tect HPV-Proofer™ assay kit.

40

10. A method according to any one of claims 1 to
9 wherein the human subjects are subjects previously
identified as infected with human papillomavirus DNA,
5 preferably in the cell or tissue under test.

11. A method according to any one of claims 1 to
10 wherein the human subjects are subjects having a
previous diagnosis of ASCUS, CIN 1 lesions or
10 condyloma.

12. A method according to any one of claims 1 to
10 when used for primary screening of individuals who
have no previous diagnosis of cervical abnormalities
15 by cytology.